

Article
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09/381286

420 Rec'd PCT/PTO 21 SEP 1999

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PCT/EP98/01653
15404P WO
attachment 1

New Claims

1. Process for isolating a purified eukaryotic proteasome preparation comprising the steps:
- (a) production of a crude extract by lysing eukaryotic cells,
 - (b) separation of insoluble components from the crude extract,
 - (c) chromatographic separation into fractions by means of an ion exchange medium,
 - (d) testing the fractions obtained in step (c) and collecting the active fractions,
 - (e) chromatographic separation over hydroxyapatite,
 - (f) testing the fractions obtained in step (e) and collecting the active fractions,
 - (g) concentrating the pooled fractions,
 - (h) chromatographic separation over a gel filtration medium and
 - (i) testing the fractions obtained in step (h) and collecting the active fractions,
- wherein
each testing of the fractions in steps (d), (f) or/and (i) comprises two determinations of the proteolytic activity one of which is carried out in the absence and the other in the presence of a proteasome inhibitor.

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2. Process as claimed in claim 1,
wherein
yeast cells are used.
3. Process as claimed in claim 2,
wherein
lactacystin is used as the proteasome inhibitor.
4. Process as claimed in ^{claim 1} ~~one of the claims 1 to 3~~,
wherein
at least one of the chromatographic separation
steps is carried out in a FPLC system.
5. Process as claimed in ^{claim 1} ~~one of the claims 1 to 4~~,
also comprising the crystallization of the purified
proteasome preparation.
6. Purified eukaryotic proteasome preparation
obtainable by the process as claimed in ^{claim 1} ~~one of the
claims 1 to 4~~.
7. Purified eukaryotic proteasome preparation as
claimed in claim 6 in a crystallizable form.
8. Purified crystallized eukaryotic proteasome
preparation,
wherein
it allows a crystallographic analysis at a
resolution of 0.28 nm or higher.

9. Purified crystallized eukaryotic proteasome preparation as claimed in claim 8, wherein it allows a crystallographic analysis at a resolution of 0.24 nm.
10. Preparation as claimed in claim 8 or 9, wherein the crystal contains a proteasome inhibitor.
11. Preparation as claimed in claim 10, wherein the inhibitor is a tripeptide aldehyde or lactacystin.
12. Preparation as claimed in ^{claim 6} ~~one of the claims 6 to 11~~, wherein it contains a proteasome from a yeast.
13. Preparation as claimed in claim 12, wherein it contains a proteasome from *Saccharomyces cerevisiae*.
14. Preparation as claimed in ^{claim 6} ~~one of the claims 6 to 13~~, wherein it contains a complex of 28 subunits which contains two molecules each of 7 different α type subunits and 7 different β type subunits.

15. Use of the purified eukaryotic proteasome preparation as claimed in ^{claim 6} ~~one of the claims 6 to 14~~ to identify and isolate new proteasome inhibitors.

16. Use of data from the crystal structure of crystallized eukaryotic proteasome preparations as claimed in ^{claim 8} ~~one of the claims 8 to 14~~ to identify and isolate new proteasome inhibitors.

17. Use of crystal structural data from the region of the proteasome pockets S1 of the subunits $\beta 1$ /PRE3, $\beta 2$ /PUP1 or/and $\beta 5$ /PRE2 to identify and isolate new proteasome inhibitors.

18. Use as claimed in ^{claim 15} ~~one of the claims 15 to 17~~ in a computer-aided modelling programme.

19. Use as claimed in claim 18, comprising a step of homology modelling in which the crystal structural data of a yeast proteasome are modified with amino acid sequences from the human proteasome.

20. Process for providing new proteasome inhibitors, **wherein**

compounds are identified based on data from the crystal structure of crystallized eukaryotic proteasome preparations as claimed in ^{claim 8} ~~one of the~~

~~claims 8 to 14~~ which have a three-dimensional structure which is complementary to the proteasome pocket S1 of the subunits $\beta 1$ /PRE3, $\beta 2$ /PUP1 or/and $\beta 5$ /PRE2.

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